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Advances in Spirulina Cultivation: Techniques, Challenges, and Applications

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Abstract

Spirulina is a microalga recognized for its nutritional benefits and its potential in sustainable food production. Existing large-scale cultivation produces spirulina of very different quality, taste, and odor. The reason lies in various approaches to the production, which range from the low-technology simple systems to high-end high-quality production for more demanding consumer market. In this chapter, we present challenges and possible solutions to ensure production of high-grade spirulina. We describe the design and crucial demands that have to be assured in the production system. The quality and productivity can be further increased by applying a bioprocess engineering approach based on modeling of the cultivation. Thermal modeling is also presented as an approach to optimize cultivation in the greenhouse systems. A spirulina production in Italy is showcased to pinpoint challenges of spirulina production in Europe. We conclude with an extensive study of regulatory framework for the spirulina production that must be taken into account for the successful algae production.

Keywords: spirulina, large-scale production, food, high-grade quality, algae production

1. Introduction

The commercial production of spirulina is well-established worldwide. In fact, spirulina is the most extensively cultivated microalga in Europe with over 200 facilities generating almost 150 tons of dry biomass annually [1]. Worldwide production in 2019 was evaluated by FAO as 56,208 tons [2]. The global spirulina market size reached € 533 million in 2023. Looking forward, IMARC Group expects the market to reach €1189.6 million by 2032, exhibiting a growth rate (CAGR) of 9.33% during 2024–2032 [3].

Nevertheless, its potential in Europe remains largely untapped as its cultivation typically takes place in tropical or semitropical regions favorable for spirulina growth.

Spirulina is a common name for commercial strains of cyanobacteria species *Arthrospira platensis* and *A. maxima*, also known by the old genus name *Spirulina* or recently even *Limnospira* (phylum Cyanobacteria) [4, 5]. While taxonomists name species according to their genetic relations as new data are discovered, technologists prefer to use common names that remain stable over time as consistency is needed in practical applications. Spirulina has long, thin filaments that are typically arranged in a spiral or helical shape, which is its distinctive feature (**Figure 1**). The helices can be tight or loose, depending on the environmental conditions. The cells are cylindrical and quite small, usually about 2 to 8 μm in diameter, but filaments can be up to several hundred μm long. The intense blue-green color of spirulina is due to the presence of chlorophyll and phycocyanin.

Spirulina has high intraspecies diversity, independent of phylogenetic affiliations or geographical locations, indicating significant physiological and metabolic plasticity [5]. This genetic variation underpins its physiological and metabolic flexibility, essential for its wide range of applications.

Spirulina thrives in alkaline environment (pH range 9.5–11) and prefers moderate to relatively high temperatures. Optimal growth conditions reported for spirulina are in the range of 300–500 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ and 25–35°C [6, 7]. However, it can also tolerate a wide range of conditions, which contributes to its widespread distribution.

Cultivated worldwide, spirulina is used as a dietary supplement or whole food ingredient. It is very rich in proteins and antioxidant compounds. Spirulina is used for the extraction of pigments such as phycocyanin, a blue photosynthetic pigment which is used in health, cosmetics, and food applications [1]. It is also used as a feed supplement in the aquaculture, aquarium, and poultry industries. Spirulina contains numerous essential nutrients, like B vitamins (thiamine, riboflavin, and niacin), and dietary minerals, such as iron and manganese [8].

The goal of this study was to address the challenges associated with large-scale spirulina cultivation and to provide guidance on producing high-grade biomass for discerning markets. Our approach utilized bioprocess engineering to achieve high-quality cultivation through enhanced environmental control in greenhouses, improved design and operations, and optimized culturing procedures. The study concludes with a detailed examination of the regulatory frameworks essential for successful spirulina production.

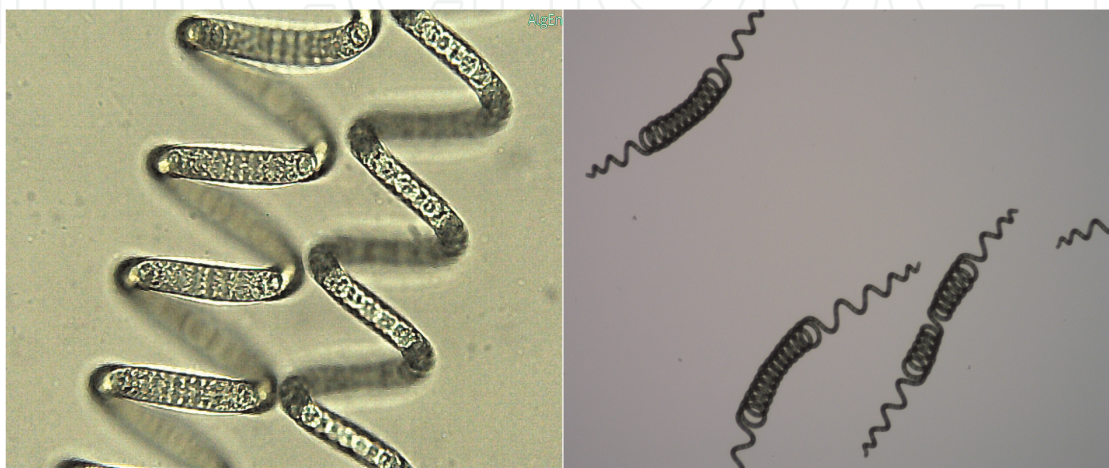


Figure 1.
Spirulina with its distinctive spiral shape.

2. Cultivation approaches, challenges, and solutions for high-grade production

Flourishing in extreme conditions characterized by high pH levels and temperatures, spirulina cultures can be very resistant to contamination. Consequently, most of the producers prefer open ponds (83% of companies in EU [1]) due to their significantly lower cost compared to photobioreactors. Pond systems consist of a raceway pond where water is mixed with a paddlewheel (**Figure 2**). They can be installed outdoors, meaning they are open to the environment, utilizing natural sunlight for algae growth, or can be housed in greenhouses for better control of the environmental conditions and prevention of infections. There are several general approaches to cultivation of spirulina for food, including (i) low technology simple systems, (ii) industrial style production in open ponds for middle quality and high-volume biomass, (iii) high-end high-quality system addressing the new-age consumer market. Each approach represents different biomass safety and quality. This chapter will present approaches for high-grade spirulina production.

2.1 Main challenges

Several factors lower the quality of current spirulina production. Open ponds offer no defense against contamination from dirt, insects, or animal remains, exposing spirulina to various environmental elements. The only form of protection employed is the regulation of pH levels to deter foreign species.

Spirulina's quality is sometimes criticized due to concerns over contaminants. Heavy metals, such as arsenic, lead, and mercury, can be absorbed by spirulina from the environment, especially when grown in open ponds subjected to pollution.

Bacterial contamination and the presence of cyanotoxins are also of concern, as spirulina is often cultivated in environments conducive to the proliferation of various microorganisms, including harmful bacteria and cyanobacteria.

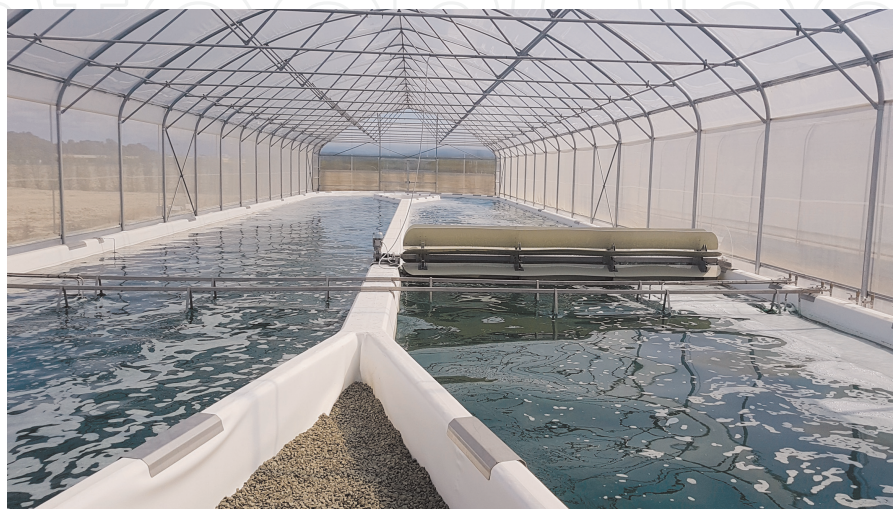


Figure 2.
Spirulina production in raceway pond in a greenhouse (Grosseto, Italy; Source: Algen archive).

Another point of criticism is the presence of polycyclic aromatic hydrocarbons (PAHs), which can form during the high-temperature drying process of spirulina. PAHs are known to be carcinogenic, and their presence in food products is highly regulated.

In the process of harvesting, media washing is done to purify the spirulina, but this step can also remove beneficial extracellular ingredients. Furthermore, the focus of current production is on optimizing the growth rate to increase the yield, rather than enhancing the quality of the spirulina.

The design, safety, and quality aspects of large-scale spirulina cultivation is thus critical for ensuring a successful operation. The design focuses on creating a controlled environment that maximizes spirulina growth while minimizing contamination risks (**Figure 2**). Safety measures include maintaining high cleanliness standards to ensure the product is free from contaminants like heavy metals and bacteria. Quality is achieved through a combination of design decisions such as closed ponds, uniform mixing, and low-temperature processing. These components work together to produce high-grade spirulina that is safe, of high quality, and produced efficiently on a large scale.

2.2 Pond systems for high-quality spirulina cultivation

High-grade spirulina cultivation should be meticulously undertaken with carefully designed ponds. Ponds need to be either covered or fully enclosed, preferably with insect nets, to shield spirulina from external contaminants such as dust, insects, and bird droppings. The pond bottom should be constructed with high-quality materials that prevent the growth of undesirable bacteria and facilitate easy cleaning.

The shape of the pond should be designed to prevent the formation of eddies, thus ensuring uniform spirulina growth and reducing energy consumption. The flow within the pond must be evenly distributed, a task accomplished by installing vanes or deflectors that spread the flow across the entire length of the pond, ensuring all parts receive equal amounts of nutrients and light. The inclusion of slanted walls can enhance wave behavior, helping to prevent stagnation and promote uniform cultivation conditions.

Regular cleaning should be conducted to maintain a pristine environment for the spirulina. Temperature regulation is critical; in hotter climates, forced air evaporation cooling systems are recommended to maintain an optimal growth temperature, while in cooler climates, heating can be effectively managed with immersed heat exchangers to avoid contamination.

Moreover, to prevent oxygen build-up, which can stress spirulina cells, a stripping sump should be included in the pond design. This will ensure that oxygen levels are balanced, promoting healthy growth and preventing oxidative damage.

Adhering to these design principles should ensure the cultivation of high-grade spirulina, yielding a product that is both safe and nutritionally rich for the consumer market (**Figure 3**).

The piping system is essential for maintaining the integrity and cleanliness of the culture environment in spirulina cultivation. Key design decisions include rigorous post-use washing of all media pipes to remove remnants and prevent contamination, strategic valve placement near the ponds for better nutrient flow control and reduced contamination risk, and the implementation of an agitation system to keep growth media in motion, addressing stagnant media issues, maintaining media quality, and preventing pipe blockages.

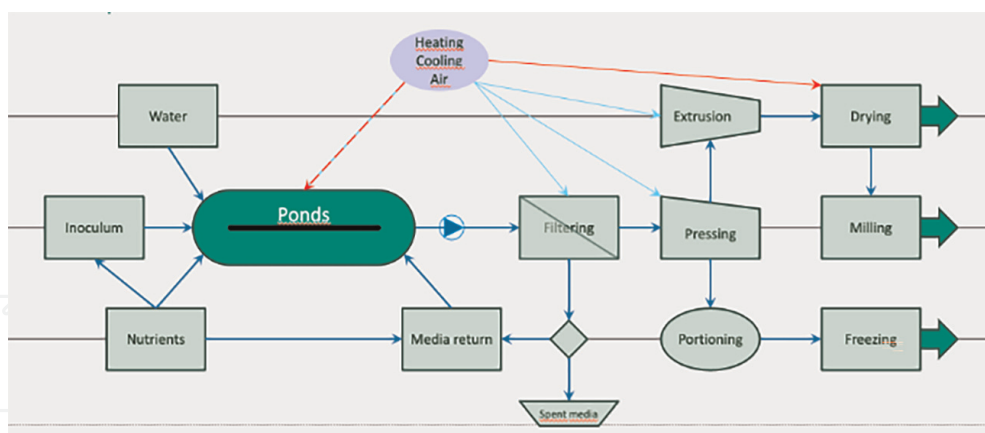


Figure 3. Top-level schematic illustrates the comprehensive workflow for spirulina production, from inoculum and nutrient preparation through various processing stages like filtering, pressing, drying, freezing, milling, and extrusion. This schematic underscores the importance of a systematic approach to spirulina cultivation and processing, highlighting the necessity of careful planning in each stage to ensure the highest quality of the final product. The inclusion of media return and spent media management also indicates the sustainability considerations inherent in the production design.

Comprehensive spirulina cultivation strategy is thus based on maintaining a clean and controlled environment to ensure high-quality production. The pH level is maintained above 10.5 to reduce growth rate slightly (in comparison to pH 9.5) but increase safety, with meticulous monitoring and adjustments using NaOH or KOH (depends on the further use of spent media for fertigation). Sufficient cleanliness is achieved with covered ponds, stringent entry protocols, and rigorous pest control measures. The system is designed to minimize stagnant culture areas and ensure all water used is sterilized (UVC at 5.3 Wh/m³), filtered, and deionized to remove most of the cations like, Ca and Mg, to prevent precipitation and white or brown flakes in biomass. Continuous Cleaning-In-Place (CIP) procedures, including post-use washes (bleach daily), washing paddlewheels and pond edges, brushing the pond lining, and regular sanitation, are enforced. The operational protocols include microscopic examinations, toxin checks, and genetic monitoring to ensure spirulina's health and safety.

2.3 Processing of biomass

Spirulina processing occurs in a controlled clean room environment to prevent contamination. For the filtering process in high-grade spirulina production, it is essential to conduct filtering at low temperatures, specifically 5–10°C, to minimize the risk of biomass degradation. The system avoids the use of water chilling and/or heat exchangers to prevent the risk of stalled biomass, ensuring the integrity of the spirulina during the filtration process. Implementing these measures within a clean room environment further ensures the purity and quality of the spirulina, safeguarding it from potential contaminants.

The process of dehydration is an essential practice for the preservation of food products over an extended length of time. In addition to reducing the development of germs, it also slows down other processes that cause deterioration. Agricultural goods undergo negative structural, textural, and biochemical changes as a result of traditional drying procedures, which leads to a significant reduction in the sensory qualities and nutritional value of the products [9]. However, drying is still an effective

method for extending the amount of time that these products may be stored effectively. The fact that this is the case has a severe effect on the quality of heat-sensitive food products that have a high nutritional content, such as spirulina. Furthermore, the selection of drying procedures has a significant influence on the overall energy consumption as well as the manufacturing cost of products [10, 11].

The process of drying spirulina is responsible for around 30% of the total expenditures incurred throughout the production process [12]. Freeze drying (FD) (also known as lyophilization), atmospheric drying (AD), vacuum drying (VD), spray drying (SD), and typical hot air drying are the procedures that are utilized the most often for the commercial application of microalgae [12]. Spirulina and other heat-sensitive cyanobacteria are susceptible to the postharvest treatment of freeze drying, which is commonly considered to be a successful method. According to Marques and Freire [13] and Oliveira *et al.* [14], this technique reduces several changes that occur to the nutritional, sensory, and physicochemical qualities of the components, which ultimately results in lyophilized products that are very similar to fresh biomass. However, in comparison to other popular drying processes, such as normal air drying, which is a more cost-effective method [15], freeze drying requires a significant amount of additional energy consumption as well as expensive equipment. On the other hand, vacuum drying provides a number of significant advantages in comparison to the conventional atmospheric drying method. These advantages include a quicker drying rate and a processing environment with lower levels of oxygen and other gases. According to Šumić *et al.* [16] and Wu *et al.* [9], these characteristics offer a significant contribution to the preservation of the quality and nutritional content of the dehydrated goods while simultaneously decreasing the expenditures that are connected with them. The spray drying process results in a high operating temperature of about 180°C, which has a detrimental impact on the quality of the dried spirulina microalga biomass. Agustini *et al.*'s work [17] suggests this is due to the fact that the heat-sensitive and essential components experience high levels of deterioration at temperatures of this magnitude.

The drying process is critical for preserving the nutritional quality of spirulina. Drying microalgal biomass is an essential process that enables the storage, processing, and transportation of the raw material. However, drying is a highly energy-intensive process that significantly impacts the ultimate structural and nutritional properties of the end product. According to Papadaki *et al.* [18], the wet spirulina biomass exhibits the maximum concentration of pigments and antioxidant activity, but a notable decline in bioactivity is found in the dry samples. Accelerated solar drying (ASD) demonstrated superior performance in the recovery of phycocyanin, whereas vacuum drying (VD) yielded a greater quantity of total carotenoids. In addition, the ASD process exhibited a greater environmental imprint across all categories, whereas the cultivation and harvesting phase of VD prior to drying demonstrated an exceptionally high carbon and energy footprint. The biomass acquired following VD exhibited a low concentration of phycocyanin, necessitating an increased feedstock quantity to yield the 1 kg of phycocyanin designated as the functional unit in the life cycle assessment. The environmental impact of phycocyanin production will be considerably diminished when one considers that the environmental footprint of microalgae production can be attributed to other products as well, including total carotenoids, chlorophylls, antioxidant compounds, and the polysaccharides of the microalgae themselves.

The decision to avoid spray dryers, which can cause high-temperature loss of essential spirulina properties (ESP), is a significant one. Instead, the production process utilizes a warm air dryer operating at 40–45°C, ensuring quick drying within 2–

3 hours. This method, combined with the use of a belt with non-sticking mesh or trays, effectively maintains spirulina's nutritional integrity. The air used in the process is dehumidified (chilled to 5°C) before being warmed to 40–45°C, with a multistage belt dryer or stacked net used to conserve space. This process also takes place in a clean room to ensure the highest quality and purity of the final product.

Stramarkou *et al.* [19] compared four methods of drying *Spirulina platensis*—atmospheric, freeze, vacuum, and accelerated solar—and found that the vacuum drying was the most effective in recovering the carotenoid content and to accomplish the quickest reduction in moisture content. Although atmospheric drying is considered an optimal technique for the preservation of phycocyanin and phenolic compounds, its protracted dehydration period renders it unsuitable for industrial implementation. Although freeze drying proved to be the most effective method for recovering β -carotene, it entails significant fixed and operating expenses. Biomass desiccated *via* solar acceleration exhibited the greatest antioxidant activity, despite the fact that a substantial degradation of the diverse bioactive compounds under investigation took place.

Packing the spirulina soon after drying and ensuring it is hermetically sealed are crucial steps for preserving its quality. The storage of the packaged product occurs in a clean area—grade 2, further emphasizing the importance of maintaining a contaminant-free environment. For fresh spirulina, packing takes place in a filter room, ensuring that the freshness and nutritional value are locked in immediately after processing.

To ensure the purity of the air within the production facility, the inlet air undergoes two-stage filtration to remove dust and microparticles, including a coarse filter followed by a HEPA filter that guarantees 99.99% filtration efficacy. The air is chilled and dehumidified to specific conditions (10°C and 9 g H₂O/m³ for the filter room; warmed to 40°C and RH 18% for the dryer room) to support the spirulina processing requirements. Moreover, air handling includes overpressure in clean areas and specific flow rates, alongside small side passes of chilled and warm air for different rooms, to maintain optimal environmental conditions for spirulina processing.

3. Optimizing cultivation: modeling

Besides the technological constraints, attention should also be paid to the effect of operating variables on the biomass viability and productivity. The selection of the proper cultivation method is pivotal for a successful production, with harvested biomass concentration and productivity standing as key parameters for such a target. A first discussion should be focused on the cultivation mode. Batch cultivation in open raceways ponds represents one of the most widespread techniques within the industrial microalgae framework, with the major advantages of reduced capital and installation costs [20]. However, such strategy does not allow to maintain stable harvested biomass concentrations and productivities, with the self-shading phenomenon and nutrient variability acting as the first causes in prolonged cultivations like these [21]. The adoption of continuous approaches should be considered in order to stabilize biomass productivity and composition: in fact, once the residence time (the ratio between the reactor volume and the inlet flow rate) is set, a steady state is naturally established, with an obtained biomass productivity and quality constant over time [22]. Continuous operation can be done by stabilizing the inlet flow rate to keep constant the residence time (chemostat) or by selecting a set point of biomass

concentration in the reactor, which is monitored by a turbidimeter and kept constant with a flow rate control, thus changing the residence time (turbidostat). However, due to technological limitations in the downstream and resulting increased initial investments for control devices, continuous operation is not yet exploited, not even at the pilot scale [23]. For these reasons, the semicontinuous cultivation mode is the most consolidated one at larger scale so far, based on removal of a certain amount of culture volume at discrete intervals, to partially harvest the biomass and replete nutrient supply. Semicontinuous operation mode at larger scale could however be improved by the knowledge acquired by lab continuous experience. As demonstrated before, the main operative condition affecting algal productivity and composition is the residence time. This applies also to semicontinuous cultivation, where the frequency and amount of harvested volume corresponds to an average residence time, according to Eq. (1):

$$\tau_{av} = \frac{\text{volume of the reactor}}{\text{volume removed/time}} \quad (1)$$

Thus, also in semicontinuous practice, the residence time should be properly adjusted to adjust biomass composition and improve productivity. Both the chemostat and turbidostat mode rationales can be applied with this perspective. The feasibility of spirulina cultivation under an optimized semicontinuous rationale was proved in the work of Pastore *et al.* [24], where *A. platensis* growth performances were tested on a 3.4 m³ pilot-scale PBR adjusting the harvesting frequency to optimize harvested biomass concentration as well as inner composition, with particular focus on protein accumulation. Thus, the investigation of the effect of operating variable at lab scale is still a powerful source of information that can be translated into good practices at larger scale, if coupled with modeling and process simulation approaches. Research at laboratory scale can be carried out to optimize the overall process performances by acting on the operating conditions. One of the aspects of interest is the effect of uncoupling the solid retention time (SRT) and the hydraulic retention time (HRT), which represents a good strategy in this perspective, as demonstrated by Barbera *et al.* [25]. Indeed, this study shows the former as the key parameter for controlling the biomass concentration within the reactor, to achieve a proper light attenuation profile according to the incident light, and thus working at the compensation point, which is the optimal one to increase the photosynthetic efficiency. On the other hand, this biomass concentration is often low, with strong impact on the water to be supplied. Thus, the process configuration for SRT < HRT is the most interesting one, as it allows to recycle the culture medium when the maximization of biomass production is the aim of the system. In this way, it is possible to meet both process performances and sustainability; the reduction of the SRT allows to stay as close as possible to the optimum value of specific light supply rate, thus benefiting in terms of biomass productivity, while the increase of the HRT minimizes the input of water and nutrients to the process, as this configuration accounts for at least a partial medium recycling. From an operational perspective, the recovery of the medium introduces a third variable to the process, namely, the recycling ratio (R); defined as the proportion between the recycled flow rate and the integrating hold up one, it can be accordingly retrieved from the selected recovered medium percentage.

The necessity of finding the optimal process operative conditions can benefit from mathematical models: being able to reduce the actual processes in the form of mathematical equations allows to produce virtual cultivation forecasts, adjustable on the

selected operating conditions, namely, the inputs given to the models themselves. Moreover, the development of mathematical models covers an important role in the overall bioprocesses design path, as it represents an essential step for scaling up operations. For a successful buildout, along with the selection of the independent material balances of the process, the correct definition of biomass kinetics must be pursued; indeed, a compromise between accuracy and simplicity must be found, usually achieved by picking the most important factors affecting such phenomenon. In mathematical terms, the most used models account for the effects of temperature, light, and nutrients availability, which are included in specifically tailored corrective factors, to reduce the species maximum specific rate of growth μ_{max} as in Eq. (2):

$$R_X = \mu C_X = [\mu_{max} \varphi(T) f(I) f(C, N, P) - \mu_{e, max} f_{maint}(I)] C_X \quad (2)$$

To account for the effect of operating temperature $\varphi(T)$, one of the most consolidated functions is the cardinal temperature model with inflection [26], in which the operating value is compared with the maximum T_{max} , minimum T_{min} and optimum T_{opt} species temperatures, as reported in Eq. (3).

$$\varphi(T) = \frac{(T - T_{max})(T - T_{min})^2}{(T_{opt} - T_{min}) [(T_{opt} - T_{min})(T - T_{opt}) - (T_{opt} - T_{max})(T_{opt} + T_{min} - 2T)]} \quad (3)$$

Concerning the light effect, several models are available [27, 28], but all should include the self-shading effect due to light absorption by biomass: this phenomenon produces an exponentially decreasing trend for light availability along the reactor depth coordinate z , commonly described by the Lambert-Beer law. Here, we present, as an example, the modified Haldane model representing the $f(I)$ in Eq. (4) [12]:

$$f(I) = \frac{1}{L} \int_0^L \frac{I(z)}{I(z) + K_I \left(\frac{I(z)}{I_{opt}} - 1 \right)^2} \text{ with } I(z) = I_0 \exp(-k_a C_X z) \quad (4)$$

The nutrients availability dependency (mainly regarding carbon, nitrogen, and phosphorus), mostly described with respect to the most limited one, is usually modelled according to the Monod-like kinetics, reported in Eq. (5).

$$f(C_i) = \frac{C_i}{C_i + K_i} \quad (5)$$

Nevertheless, this model has a strong limitation, as it considers a fixed biomass on nutrient yield. For this reason, Droop model should be applied: indeed, by introducing the concept of limiting nutrient quota, namely, the amount stored within biomass, a more accurate description of microalgal uptake dynamics can be achieved [29].

Finally, the maintenance is added to the overall dynamics, with the maximum maintenance energy $\mu_{e, max}$ adjusted according to the light availability correction factor $f_{maint}(I)$ [30] in Eq. (6).

$$f_{maint}(I) = \frac{I_0}{I_0 + k_{I, m}} \quad (6)$$

Eq. (2), namely, the standard microbial kinetics, may be further modified by introducing other factors that can affect growth performances. For instance, the presence of some organic metabolites may inhibit cyanobacterial growth, which must be accordingly modeled. This is the case when a medium recycling configuration is adopted: indeed, inefficiencies in the harvesting system may lead to an ineffective separation of such compounds, which may then accumulate within the reaction environment. Specifically for *Spirulina* sp., the literature reports exopolysaccharides (EPS) and free fatty acids as the main contributors in relation to this. If, on one hand, the active inhibitory effect of fatty acids accumulation is taken for granted [31], the inhibition mechanism provided by EPS is still subjected to debate. For instance, some works sustain EPS active role in *Arthrospira platensis* growth inhibition, while others support a more indirect role, with EPS accumulation increasing medium viscosity and negatively affecting the subsequent biomass harvest: this may induce a reduction in filterability, leaving room for the buildup of inhibitory compounds within the reactor environment. For example, see [17, 32]. Regardless of the rationale, this confirms that EPS may be seen as the key component while accounting for a potential inhibition correction of biomass kinetics. These experimental findings could be beneficial in view of process optimization if accounted by proper modeling techniques, aimed at tracking the system performance while simultaneously keeping in mind such inhibitory phenomenon; this way, the degree of medium recovery up to a value such that the EPS concentration with the reactor environment can be kept under control.

4. Thermal modeling of raceway ponds for microalgae cultivation

4.1 The thermal model for the greenhouse-pond system (GPS)

Mathematical growth models have been recently developed to forecast the performances of microalgae cultivation systems and the consequent cost-effectiveness of temperature control schemes (see Section 3). Since temperature is a crucial input parameter, a proper thermal modeling is of great aid to the simulation accuracy. Although validated models for open air cultures are already available [33], thermal evaluations under greenhouse (GH) are less well-established; nevertheless, a GH is typically needed to limit exogenous contamination to produce medium to high-quality biomass.

The dynamic greenhouse thermal model taken as reference was developed by Li *et al.* and describes shallow ponds for aquaculture purposes covered by a GH equipped with two cover layers [34]. It is a mechanistic conceptual/gray-box model, based on the modeling of the major heat exchange mechanisms and heat/mass balances across each component of the system. The model (**Figure 4**) considers perfectly mixed/homogeneous layers in the three spatial dimensions as for the external air, the air between the covers (when present), the GH internal air, and the pond water so that model components include: (i) the external ambient air (e), (ii) the air between the external (c1) and internal (c2) covers of the inflated double-layer glazing, (iii) the greenhouse (GH) inside air (i), and (iv) the raceway water (w). The raceway is assumed to cover the whole GH floor, and the soil beneath the raceway (s) is assumed to have a vertical temperature gradient until the isothermal layer is reached, whose temperature is a Dirichlet boundary condition, and it is considered equal to the annual average value of the external air temperature profile.

The original model was adapted to raceway pond (RWP) configurations, and some extensions were made [35], such as: (i) the development of the model for single-cover

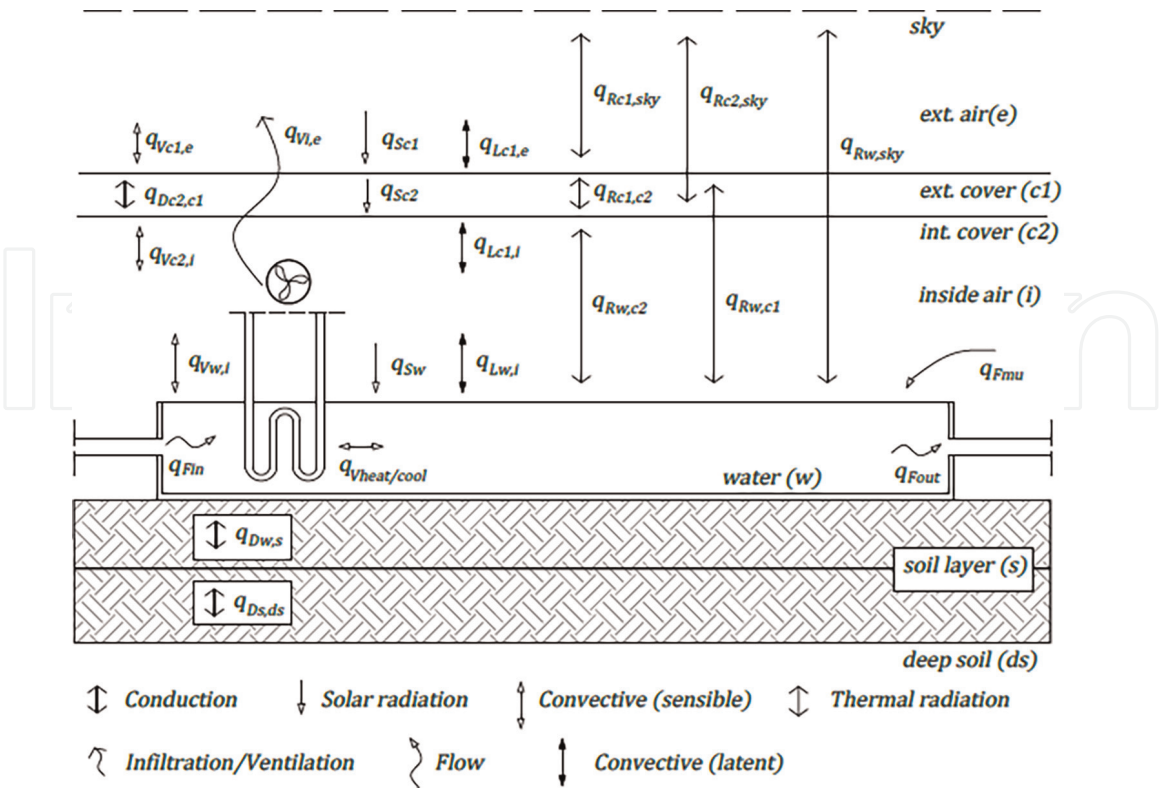


Figure 4.
 Layers of the GHP system and heat fluxes for a double-cover GH.

(c) configuration, (ii) the presence of inflow/outflow and make-up water, (iii) the presence of RWP insulation, (iv) the possibility to apply temperature control strategies (water heating/cooling, mechanical ventilation), and (v) the impact of GH cover on the penetration of the photosynthetic active radiation (PAR).

4.1.1 Model structure

The overall differential algebraic equation (DAE) system is defined as in Eq. (7), where x are dynamic variables, z are algebraic variables, u are control inputs, θ are fixed parameters, d are external disturbances, and y are measurable outputs. Functions f and g are, without loss of generality, nonlinear (and in some cases $\in C^0$).

$$\begin{aligned} \dot{x} &= f(x,z,u,\theta,d) \\ 0 &= g(x,z,u,\theta,d) \\ y &= h(x,z,u,\theta,d) \end{aligned} \tag{7}$$

The state variables are three temperatures (T) of the interacting heat capacities and the water vapor content $e_{int,air}$ (kg m_{air}^{-3}) of the greenhouse internal air. The state variables interact with each other *via* heat fluxes Φ (W). The dynamic equations describe the energy balances for each uniform layer, that is:

$$CVdT/dt = Q(T_{in} - T) + \Phi(T,\theta,d) + u \tag{8}$$

where C ($\text{J m}^{-3}\text{C}^{-1}$) is the volume-specific heat capacity and V (m^3) is the volume.

The first additive forcing term is related to the enthalpic contributions of inlet/outlet flows, where Q ($\text{m}^3 \text{s}^{-1}$) is the flow rate, which are present only for the internal air and for the raceway pond (RWP) water layers. Water mass balance over the greenhouse internal air is used to calculate $e_{int,air}$, which is involved in the latent-convective heat exchange. The cover temperatures are algebraic variables (negligible heat capacity). With reference to **Figure 4**, the heat exchange mechanisms are:

- *Radiation qR* . As the covers have very high transmissivity, both covers and water pond are heated up by solar near-infrared radiation (NIR) during the day and cooled by far-infrared radiation (FIR) emission to the sky during the night. The thermal radiation properties of plastic covers vary with the amount of condensate covering them.
- *Sensible convection qV* . outside air convection is primarily impacted by greenhouse geometry and wind speed, whereas inner air convection is primarily influenced by temperature differences between layers. Heat exchange with external air is also considered *via* an infiltration rate Ra (h^{-1}). Although it varies with the inside-outside temperature difference and outside wind speed, the heat loss resulting from infiltration is generally small compared to the overall heat loss, and therefore, Ra was assumed to be constant.
- *Latent convection qL* . Heat exchange for water phase transition primarily depends on sensible convective heat transfer coefficient, Lewis number, and difference in water vapor concentration as driving force.
- *Conduction qD* . Conductive exchange is primarily present between the pond and the ground, and between the internal and external air.

The enthalpic contribution given by pond make-up water was also considered. The model has a time step resolution of 120 seconds.

The PAR ($\mu\text{mol m}^{-2} \text{s}^{-1}$) that reaches the pond surface is given by Eq. (9), that is:

$$PAR_w = 2.105\tau_{S,c}^2(1 - \rho_{S,w})I_0 \quad (9)$$

where 2.105 is a conversion factor, $\tau_{S,c}$ is the transmissivity of each cover to solar radiation, $\rho_{S,w}$ is the reflectivity of the water surface, and I_0 is the outside global solar radiation (W m^{-2}).

4.1.2 Model parameters and input data

The GHP thermal model includes the following classes of parameters (θ): (i) empirical parameters subject to calibration, (ii) physical parameters (for example pond water, soil, and air thermal/optical properties), (iii) empirical parameters from literature correlations (for example sky/external air temperatures correlation), and (iv) input design parameters (for example hydraulic retention time (HRT), pond liquid height, GH geometry, and cover material properties).

As a matter of fact, compromise between model complexity, computational time, and modeling effort was made so that the model contains both conceptual correlations and 6 empirical parameters subject to case-specific calibration; these are: (i) the

infiltration rate (Ra), (ii) the thickness of the non-isothermal soil layer (Ls), (iii) the convection regime between the cover layers (k_1 multiplier coefficient of $qD_{c1,c2}$ —in case of a double cover), (iv) the reflection contributions to solar irradiance from the surroundings to the greenhouse external layer (k_2 multiplier coefficient of qS_c), (v) the convective heat transfer coefficient between water and internal air (k_3 multiplier coefficient of $hV_{w,i}$), and (vi) the convective heat transfer coefficient between internal air and internal cover (k_4 multiplier coefficient of $hV_{i,c2}$).

The inputs to the GPS model are process design parameters and weather data. Hourly weather data are required for: (i) external air temperature (Te), (ii) global solar irradiance at ground level (I_0), (iii) external air relative humidity (RHe), and (iv) external air wind velocity (ue).

4.1.3 Temperature control

The GPS model was integrated with a pond water temperature feedback control scheme (es. multiple input single output (MISO) proportional-integral-derivative (PID) control) for the estimation of: (i) the heating/cooling loads and peak powers from direct submerged heat-exchangers and (ii) the mechanical ventilation load and peak power. Mechanical ventilation was modeled with an additive term on Ra .

4.2 The integrated thermal and biological model

4.2.1 The opportunity of optimizing temperature regulation

Microalgae metabolism is particularly sensitive to temperature, and literature is rich in models that consider the effect of temperature on microalgae growth and respiration. An effective model that quantifies the temperature dependence of growth and respiration is the Cardinal Temperature Model with Inflection (CTMI) proposed by Rosso *et al.* [36] and reported in Eq. (3). It includes three parameters (the cardinal temperatures: T_{max} , T_{opt} , T_{min}), which define the optimal working range for each microalgae strain [37]. In addition, temperature also plays a role in physiochemical equilibria, such as gas/liquid exchange, solubility, and dissociations that indirectly add impacts on metabolism.

When the GPS is integrated with biological or with biological and physical-chemical models, a comprehensive understanding of the algae biomass production facility can be run leading to a proper estimation of microalgae productivity and the consequent cost-effectiveness of thermal regulations. With calibrated GPS and biological growth models, the integrated assessment can be adapted to different climatic and biological conditions, allowing to improve the techno-economic analysis (TEA) and the consequent feasibility and scalability of microalgae cultivation. Indeed, an objective function that entails both the higher revenues and the higher costs from temperature regulation can be set for scenario analysis and optimization.

4.2.2 A case-study

An example is presented in Carecci [21], where the GPS is integrated to the comprehensive biological and physical-chemical ALBA model [38]. The simulations carried out with the ALBA model considers climate conditions as they are computed from the GPS model (water temperature (**Figure 5**), evaporation ($m^3 day^{-1}$), and light intensity ($\mu mol m^{-2} s^{-1}$)). The case study considers a biorefinery located in

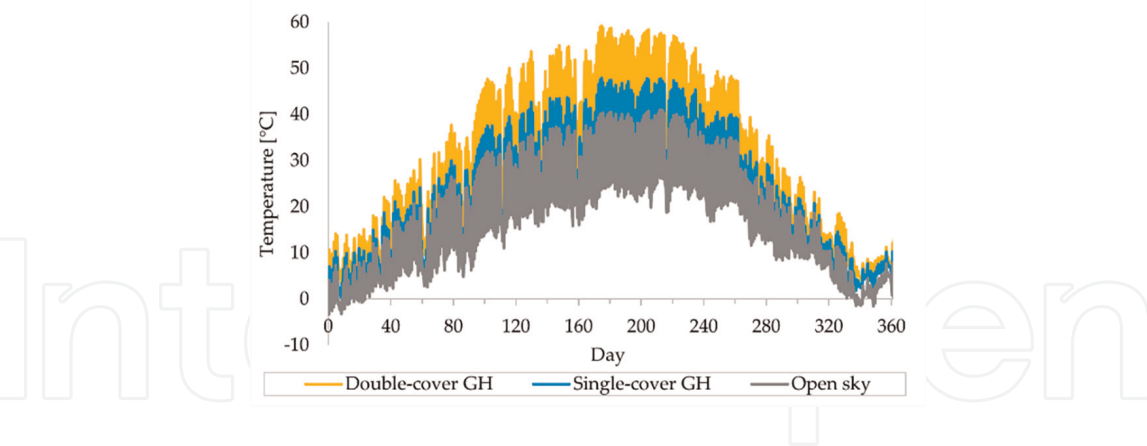


Figure 5. Yearly RWP water temperature dynamics for open, single-covered, and double-covered GH configurations.

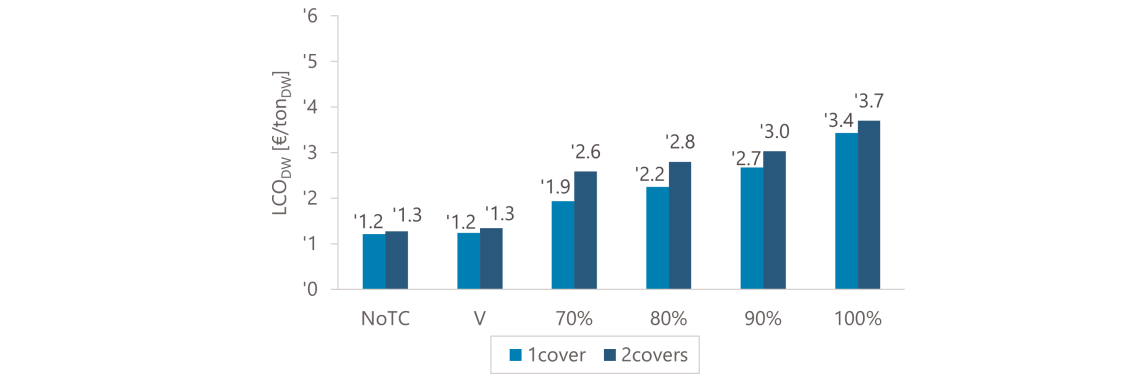


Figure 6. Levelized costs of microalgae biomass production (LCO_{DW}) for different covering, control strategies, and set points scenario.

Lombardy (northern Italy) for the cultivation of a *Chlorella-Scenedesmus* consortia for biostimulant production on agrozootechnical liquid digestate. In that case study, different scenarios were evaluated considering different GH covering alternatives, water temperature set points, and temperature control strategies. The latter were selected by assuming different temperature ranges around the optimal value as suggested by the CTMI curve for that microalgae community, which would be allowed in the pond by the T-controller. The productivity computed from the ALBA model was combined with heating/cooling loads provided by the thermal control logic in a comprehensive economic framework, where levelized costs (**Figure 6**) and return of investments were evaluated. For the specific location and market conditions of the case study, the best design option was to implement a single-cover GH, regulated only by summer cooling *via* both mechanical ventilation and water-cooling. Similar simulations can be easily extended to spirulina production by adapting the biological model parameters to describe spirulina growth and respiration rates.

5. Upgrading the biomass quality

Besides the improvements on growth rates by applying different cultivation strategies (see sections above), biomass quality improvements in terms of its biochemical composition (proteins, carbohydrates, lipids, pigments, and mineral content) are also

possible by controlling some of the cultivation parameters. The main parameters that can be considered for this purpose are light intensity and quality, time of harvest, nutrient availability, and salinity of the growth medium.

Light has not only an important role on the cell growth, but it influences the biochemical composition of the biomass. It is generally observed that at higher light intensities, the biosynthesis of carbohydrates is favored, while at lower intensities, the protein and pigment content (phycocyanin and chlorophyll) is higher. This is because at increased intensities (however below levels that cause photoinhibition), photosynthesis is improved and the photosynthate is directed towards the biosynthesis of carbon and energy storage compounds like glycogen, which then is utilized further as metabolic energy carrier for biosynthesis of other metabolites, or respiration. When artificial light is applied, the intensity could be optimized for growth and protein and phycocyanin productivity. However, light intensity is a parameter that could not be efficiently controlled in cultures grown with solar energy due to the high fluctuations during the day, where typically at midday, the highest intensities occur. At very high light intensities, photoinhibition typically takes place that decreases growth and negatively influences protein and phycocyanin content [39, 40]. In practice, where production is performed with solar light, shading of the cultures is of importance to avoid photoinhibition. Since the quality of the light influences the biochemical composition of spirulina, the shading of the cultures could be performed by using colored filters absorbing most of the incident light spectrum and allowing passing the desired ones. As was reported by Kilimtzidi *et al.* [41] in small-pilot open pond experiments, shading of spirulina with red filters improved the protein and phycocyanin content. Also, the harvesting time has an important effect on the biomass quality, since it has been observed that in the early morning, the percentage of the proteins and of essential amino acids is the highest [42, 43].

Spirulina is typically cultivated with growth media with relatively high bicarbonate concentrations and total salinities (as sodium ions) to avoid any significant contamination with other microalgae or cyanobacteria. The most common growth medium used for spirulina production is Zarrouk that contains around 5.5 g-Na⁺/L. It was found, however, that the protein content of spirulina was around 11% higher when lower total sodium ions (4 g-Na⁺/L) was used [44]. Nevertheless, despite that increasing salinity negatively affects protein content, the use of seawater to formulate the growth medium could be a strategy for replacing fresh water and the production of biomass with increased unsaturated lipids (Oleic acid, Cis-9 (C18:1), and Palmitoleic (C16:1)) [44, 45].

For improved protein content, another possible strategy is the addition of small amounts of glycerol (0.5–1.5 g/L) [46, 47]. However, this strategy could negatively impact the cultures since heterotrophic microorganisms can grow when organic carbon is applied, especially in open-pond facilities where no sterile or axenic conditions can be achieved.

Spirulina contains minerals such as iron, magnesium, calcium, zinc, and so on and could be a good source when used as a food or feed supplement. The increase of these minerals in the growth medium could lead to the increase of their content as it is having been demonstrated in several studies [48–50].

Despite that spirulina has been produced with the main target in its protein and phycocyanin content, novel products could be also developed focusing on other compounds like polysaccharides. In the study of Markou *et al.* [51] under phosphorus limitation, it was found that spirulina was significantly enriched in 1.3:1.6-β-Glucans, which are considered to have antitumor, anti-inflammation, and antiviral activities.

6. Showcase Algaria, Italy

To better understand the importance of operating variables on biomass productivity and quality, a case study is presented here, to highlight the aspects that should be carefully considered to fill the substantial gaps related to scaling up and industrialization efforts of spirulina cultivation. Practical experiences from operating commercial-scale facilities highlight deviations from standardized laboratory conditions, both in terms of duration and due to biotic and environmental factors. Long-term evaluations tailored to specific production needs are thus essential for assessing the industrial scaling up of this nascent industry, posing new challenges also to the lab scale studies. One of the primary challenges faced by raceway facilities is their exposure to ambient conditions, which introduces various variables such as dust, insects, and bird droppings.

While covering the raceway with a greenhouse and protecting it with mosquito netting could mitigate these issues, this solution may impact light availability. The transparent plastic cover of the greenhouse reduces light penetration, resulting in an average reduction of 50% throughout the year, with the maximum impact observed during the summer. Despite this drawback, the greenhouse environment offers effective thermal management, providing a potential solution to address low temperatures. With a temperature difference of around 10°C between inside and outside the greenhouse during daylight hours, this setup could significantly extend the productive seasons in temperate regions by 2 or 3 months. This extended growing period has the potential to enhance overall spirulina production and contribute to the sustainability and profitability of raceway facilities. However, during the wintertime in temperate zones, temperatures can still decrease below zero, so it may still be challenging to maintain temperatures above the critical threshold of around 15°C, necessary for maintenance, especially for thermophilic microorganisms like spirulina.

The fluctuations during the 24-hour cycle are also important as the physiological aspects of photosynthesis and respiration are primarily influenced by light and temperature. Relatively low temperatures coupled with high light intensities—a common occurrence during summer/spring mornings in temperate zones—can lead to photo-system damage. Vonshak and Richmond [52] demonstrated that photoinhibition can occur in outdoor cultures, resulting in up to a 30% loss of biomass production rate. This limitation emphasizes the importance of exploring alternative solutions or implementing supplementary heating systems to maintain consistent and optimal conditions for spirulina cultivation throughout the year.

Algaria company in Italy addresses this challenge by utilizing heat generated by a biogas plant for spirulina production. This approach effectively reduces the daily temperature variation by $\pm 5^{\circ}\text{C}$ and ensures that temperature never falls below 15°C [19]. As a result, spirulina production flourishes, with reported yields ranging from 6 g/m² day⁻¹ in winter to 16 g/m² day⁻¹ in summer. This demonstrates the potential of innovative solutions to enhance sustainability and profitability in raceway facilities, particularly in temperate climates.

As stressed in the previous sections, it is crucial to consider the production process and operations, as they significantly influence the final production output. Operational conditions interact in complex ways; that is why an integrated approach to analysis is necessary. While continuous operation is ideal, maintaining a fixed dilution rate or biomass composition at a large scale can be problematic due to climate fluctuations and operational difficulties. A common approach is the use of a semicontinuous mode, like chemostat or turbidostat as described in Section 2. Seasonal patterns, such

as different growth rates and biomass concentrations in winter and summer, must be still considered. Challenges arise during periods of negative growth rates, often caused by factors such as low light availability and moderate temperatures, which increase organic matter consumption compared to production by photosynthesis. Key parameters affecting productivity, such as dilution rate and harvesting frequency, should be chosen based on physiological traits of the culture and the objectives of the production system. Misalignment between harvesting rates and growth rates can lead to culture collapse. Therefore, robust operational protocols and adaptive management strategies are essential to ensure consistent and efficient spirulina production in raceway facilities.

7. Regulatory framework for spirulina production

Regulatory and market demands must be thoroughly considered in the spirulina production. The spirulina market is characterized by a significant presence of small and medium-sized enterprises (SME) that produce and sell spirulina products directly to consumers (business-to-consumer approach). These companies are usually run by few people with a background in a specific sector that need to multitask in different work areas. When starting a spirulina business, it is fundamental to assess the regulatory framework affecting the different steps of the value chain. The regulatory framework can be regarded as a tree, where the different applications of spirulina products share a common root (permissions, waste management, etc.) and trunk (labor safety, equipment operation, etc.) (**Figure 7**). Spirulina regulation falls on the algae framework, and its application is sometimes complex and demanding. This section aims to provide spirulina entrepreneurs with some insights and guidance through the regulatory framework surrounding spirulina production. Most importantly, algae production in Europe is costly partly explained using complex equipment (e.g., closed photobioreactors or systems inside greenhouses) and compliance with stringent laws and standards for safety and quality. The complex regulatory landscape in the European Union is the basis for high quality and certified production of spirulina. Nevertheless, the low production of high-quality spirulina in Europe in front of imports from countries with a larger production and older tradition often results in poor quality products filling the European market. Recognition by consumers of the connection between the regulatory framework and product quality should counterbalance the spirulina market in Europe.

First of all, it is necessary to define what spirulina is from a regulatory point of view in Europe. European Standards [53] and the EABA [54] integrate cyanobacteria, microalgae, macroalgae, and Labyrinthulomycetes in the same functional group (algae) due to their similarities in functional properties and derived products. Algae and algae products fall under the regulation of aquaculture products mainly due to their cultivation in aqueous medium [55, 56], despite sharing traits of a biotechnological process (microorganism) and agricultural process (autotrophic growth). It is therefore relevant to distinguish the end stage of the final product since it will determine the hygiene regulation that must be followed. On the one side, regulations referring to agriculture (primary production) will apply when the purpose is growing biomass and harvesting. On the other side, regulations referring to industrial production (transformation) will apply when the process includes concentration, extraction, purification, and packaging. Finally, Spirulina is nowadays mainly sold as a food supplement or nutraceutical, but in the last years, it has gained interest in different

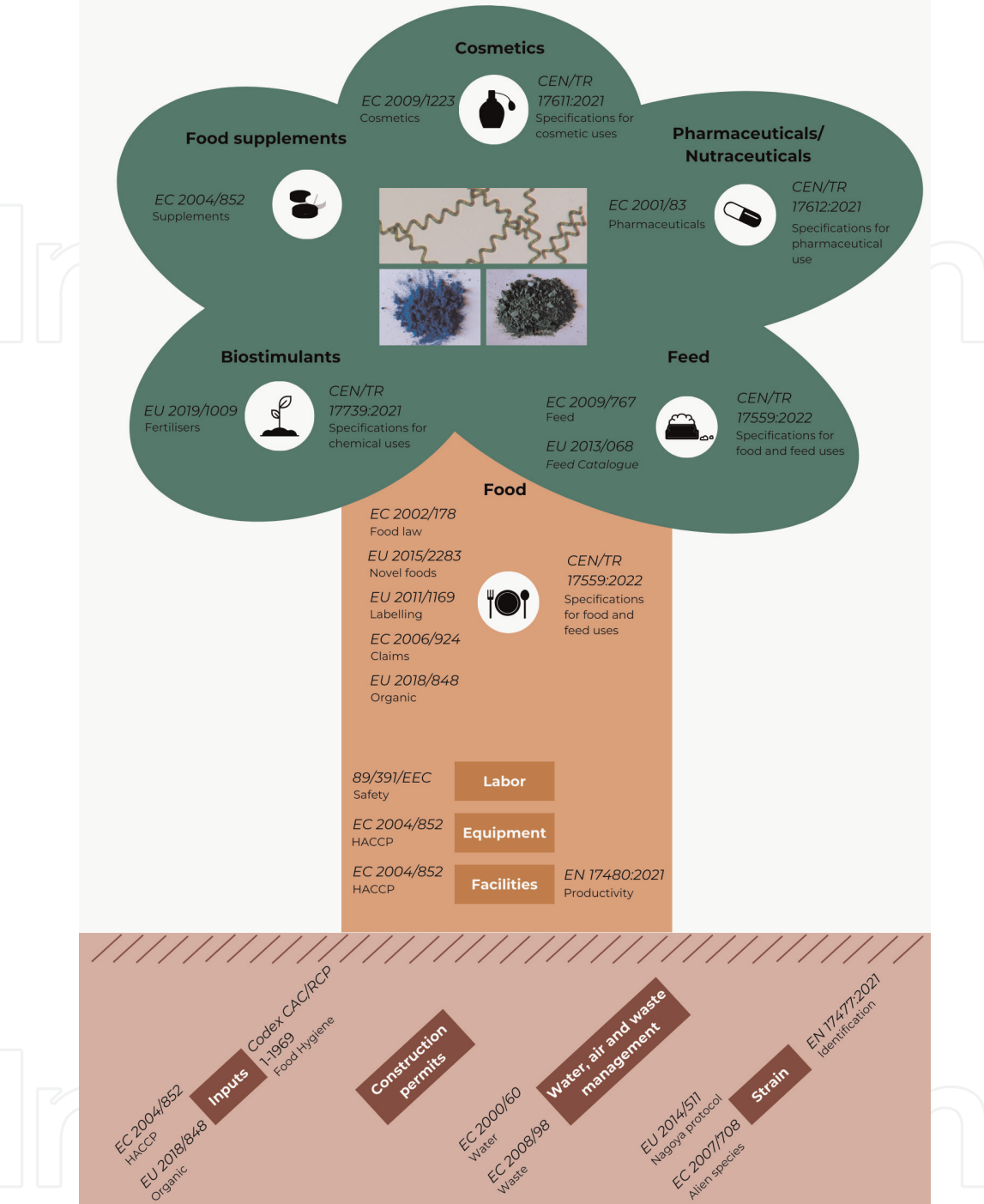


Figure 7. Overview of the regulatory framework of spirulina products with special emphasis on the law and standards applying for products for the food sector. The scheme has a tree shape, distinguishing the most relevant laws for inputs, construction permits, waste management, and strain (roots); the laws/standards applying for labor, equipment, and facilities (trunk); and the laws/standards for the different applications of spirulina products (branches).

market sectors as a feed or biostimulant. Depending on the application of the final product, there will be a specific regulation to be applied. For instance, safety of food products is regulated by EC 178/2002 [57], but food supplements additionally require the compliance with directive 2002/46/EC [58]. The feed, cosmetic, and biostimulants sectors are regulated by the 2009/767/EC [59], the 2009/1223/EC [60], and the 2019/1009/EC [61], respectively. Regarding the nutraceutical or functional foods, there is

not a specific regulation, and they may fall under those of pharmaceutical products (2001/83/EC [62], food for specific population groups (2013/609/EU [63]), or food supplements [33]. The following lines will focus on the use of spirulina for food and food supplement applications (**Figure 7**).

Globalization, innovation, and free trade have contributed to the rise of nontraditional food products across the globe. Algae-related products are one example of nontraditional food with a short history of consumption in Europe. Regulation EC 2015/2283 (formerly EC 258/97) set the boundaries and procedures for selling food products that are safe for humans and that were not consumed before May 15, 1997 [64, 65]. Therefore, products that were not present in the European market before this date are considered Novel Foods and they must be approved by following an authorization procedure. Most algal products fall under the Novel Food category, and as such, their safe consumption must be ensured. The authorized products are available in the Union List of Novel Foods, which is regularly updated (CIR EU 2017/2470) [66]. Foods that are not “Novel” are listed in the EU Novel Food status Catalog, which is a nonbinding list used for guidance purposes [67]. The presence of spirulina in the Catalog indicates that several species of spirulina were consumed in Europe before May 15, 1997.

Starting from the design phase, adequate permissions are required for the building and construction of the facilities while bearing in mind the final application of the product. Once the facility is operational, these permits must be maintained over time. The operation of the facilities requires the use of several inputs ranging from the raw materials to cultivate spirulina such as nutrients, carbon dioxide, or water to energy for culture circulation and mixing. Both the raw materials used for cultivation as well as the material of the equipment used during the process must be food grade. Finally, generated waste must be managed during the operation of the facility. Environmental regulations exist that cover all impacts of spirulina production including water [68], waste [69], and emission management. Water is the most relevant topic, and the framework is similar to aquaculture and agriculture practices. In general, EU environmental law sets principles and requirements through Directives instead of Regulations, to leave some flexibility to the member states to adopt their legislation (subsidiarity principle). On the contrary, this approach leaves stakeholders facing uneven competition and hurdles due to a lack of harmonization in the legal framework. Specially for these matters, it is therefore necessary for stakeholders to consider local regulations since it is up to the specific local competent authority to comply with EU law.

All products fit for human consumption must be safe for the consumers. The safety of spirulina is mainly determined by its purity and the absence of contaminants such as pesticides, heavy metals (EC 915/2023 [70]), or microorganisms (EC 2073/2005 [71]). The safety of the product can be ensured when the cultivation and further processing of spirulina are performed in facilities and with equipment that comply with the level of hygiene stated in EC 2002/178, and by suitably trained operators. One approach to minimize risks and ensure the quality of food product is through the application of a Hazard Analysis and Critical Control Point (HACCP) plan (EC 2004/852) [72]. Shortly, the implementation and maintenance of a HACCP plan helps in the identification of hazards, establishment of monitoring procedures and correcting measures, and documentation for tracking proper hygiene practices. Besides the HACCP system, there are other similar approaches to ensure food safety and quality such as the Good Manufacturing Practice (GMP) system. The GMP system was originally designed for safety in the pharmaceutical sector, but it is also functional in

the food sector. The Codex Alimentarius CAC/RCP 1-1969 [73] is a document developed by FAO and WHO that summarizes the principles of Good Hygienic Practices (GHP) and HACCP. Also, private certifications exist such as the Good Agriculture Practices (GAP) developed by Globalg.a.p. For food applications, the HACCP system is highly advisable in Europe to grow and sell spirulina that complies with food law regulations. Nevertheless, this system is not mandatory when spirulina is only cultivated (primary sector activity), but it is mandatory when the process includes transformation such as drying and/or packaging (secondary sector, i.e., industry). The business operators play a central role in the implementation of this system since they must follow the established hygienic measures and train operators to comply with them. In addition, business employers must comply with labor safety regulations defined in directive 89/391/EEC [74].

The quality of the product is a parameter of utmost relevance for the customer, and it is not an exception for microalgae. The quality of the spirulina products is specified in technical data sheets (TDS), Certificates of Analysis (CoA), or safety data sheets. These documents include information regarding the nutritional value, the storage conditions, and other properties of the product such as the purity. The nutritional information for consumers is described in the regulation 2009/1169 [75], which clearly states all the information and labeling rules of food products. It is also a common practice to include nutritional and health claims for food products in commercial communications. Regulation 2006/1924/EC addresses the potential health and nutritional claims that can be done based on the nutritional properties of the food product [76]. The purpose of this regulation is to ensure that claims are truthful, clear, and based on scientific evidence. At the moment, there are still no approved health claims for spirulina, despite several applications being under revision.

Different certificates of quality exist that can increase the value of the product. Certification is a proof of compliance, given by a third party ("Certification body"), between the supplier and the customer, usually released on a voluntary basis in front of Standards (reference documents). Despite being voluntary, it is yet required by the customer since it has a market impact. On the opposite, regulatory compliance is mandatory, verified by Official Authorities, and has a legal impact. Different types of certifications prove that a product has been developed according to certain quality, social, environmental, or religious standards. Certification of quality in products may also aim to protect the practices, origin, and tradition of certain products in Europe and includes different types of certificates such as Organic, Protected Denomination of Origin (PDO), Protected Geographical Indication PGI, or Traditional Specialties Guarantee TSG, as stated in regulation 2012/1151 [77]. Religious Certifications such as Kosher and Halal certify that the product was produced according to Kosher and Islamic law requirements, respectively. Environmental certificates such as ASC-MSC Seaweed Standard or Demeter Biodynamic Certification [78] identify those products whose production minimizes their impact on the environment.

Among the different types of certificates, the European Organic certification is the most adopted by spirulina producers. The Organic certification guarantees that a certain product has been produced according to sustainability principles and methods that minimize the impact to the environment, protect the biodiversity, and contribute to the local farming. In that case, a European regulation states the principles and rules (EC No 2018/848) and a certification body, which can vary between European countries, evaluate the compliance of a product to organic production methods [79].

8. Conclusions and prospects

In this chapter, we have examined some aspects of spirulina cultivation. Spirulina is probably the easiest microalga to cultivate in a safe and efficient way. Compared to other microalgae, there is ample experience in cultivation and human consumption of spirulina, but the exposition in this chapter shows that we are far from agreeing even on basic terms like safety, quality, and optimal cultivation technology, even less on engineering of spirulina cultivation, harvesting, and drying systems. Part of this disagreement is rooted in biology and enormous intraspecies diversity as well as high variability of climatic and environmental conditions. Even larger part of disagreement belongs to different objective values: what is safe spirulina, does market demand organically certified cyanobacteria, what are the quality criteria of a good product, and what are legal and regulatory constraints? In short, what is the market demand?

Europe is lagging on algal production compared to other parts of the world. Spirulina has gained some market share in European markets, although the popularity of spirulina in different markets varies significantly; there are countries with decades of tradition of spirulina consumption and countries where spirulina is virtually unknown. Most of spirulina consumed in Europe is imported from various sources; some are decently good, some less so; there is almost no criteria to evaluate product quality or product value.

Spirulina cultivation in Europe cannot compete with the cost of production elsewhere—cultivation conditions, labor, and investment cost, and also, regulatory constraints in other parts of the world are simply more favorable. However, spirulina made in Europe may and must be competitive in quality, taste, local origin, branding, product safety, and environmental impact.

Variations in product quality have already raised concerns of various consumer organizations on spirulina safety [80–82]. Up to now, most of the warnings were formulated as advice to consumers to rely on reputable sources of spirulina, but the community can easily lose credibility in the eyes of the consumers. So, adhering to strict safety and quality standards to avoid heavy metal, bacterial, or cyanotoxin contamination (in the pond or after filtering) and avoidance of higher temperatures are important for the whole community. We cannot be happy to produce safe spirulina while unsafe spirulina is being on the market—consumers are prone to generalize, and they do not distinguish different production methods.

Another important conclusion of the presentations in this chapter can be summarized with a statement “scaling-up is not easy.” Methods and techniques that work in the lab or even in a small pond are not directly applicable at large scale. Cleanliness and contamination prevention in the lab is usually a normal routine, while it is impossible to protect a large pond from all contaminants, even more, a contamination source is not easily discovered at large scale. This means that upscaling is a task of professionals with experience and methodology that will result in a manageable system and manageable processes.

To achieve consistent high quality and high productivity, process optimization should focus on both upstream and downstream operations. The design, safety, and quality aspects of large-scale spirulina cultivation are critical for ensuring a successful operation. The design focuses on creating a controlled environment that maximizes spirulina growth while minimizing contamination risks. Safety measures include maintaining high cleanliness standards, employing clean room technologies, and implementing safe procedures. Quality is achieved through a combination of design decisions to prevent contamination, ensure consistent nutrient distribution, and

maintain nutrient integrity. These components work together to produce spirulina that is safe, of high quality, and produced efficiently on a large scale. Last but not least, best quality standards and demanding regulatory framework such as those in EU have huge impact on quality and safety of EU farmed spirulina.

The regulatory framework around a specific spirulina product is thus an aspect that must be considered at early stages. Despite lack of specific spirulina production regulation or standards, it is worth noting the use of the regulatory framework of the aquaculture sector as a reference point for stakeholders. We should consider good regulation as an asset rather than an obstacle. Clarification and awareness of the regulatory framework is important for hygiene and safety of the products, the people in the process, as well as the sustainability of the whole process.

There is more to be done at the R&D level. The recycling of water and excess nutrients plays an important role in quality, sustainability, and profitability. Based on the varying experience of different producers, more R&D work is required. Another almost untouched area is the taste of spirulina: different products come with different tastes, which is of a primary importance for consumers with no explanation what is determining the taste of the product.

A very important basis for process control is good modeling that includes relevant physical, chemical, and biological factors into a powerful process models that can be used as digital twins and predictive component of the process control algorithms. Kinetic models cannot be replaced by artificial intelligence; they may be augmented by AI in some aspects like parameter adjustments. Availability of affordable metagenomic tools seems to call for inclusion of metagenomic data into the models as a verification and parameter adjustment mechanisms. Higher level of control in very large production systems demand early warning functionality that will enable proactive controls.

Spirulina farming is frequently considered as the entry level technology to a more demanding farming of other species. Provided that spirulina market exists, it is also the safest investment prior to moving into some other higher value products. Spirulina itself is a safe and sustainable source of protein, antioxidants, and other substances that will become more and more important as replacement of other protein sources with high environmental impacts. In this view, we can consider it a strategic technology, and there is no doubt Europe has to be active in its development to reduce the lag from countries where these technologies are already developed at very large scale. We have to do it considering our own climatic, environmental, and market conditions, and it seems we can do it in a commercially viable way.

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